A diagnostic assay for detecting and/or quantifying $A\beta$ peptide which may be present in a candidate solution, comprising:

- contacting the candidate solution with a solid support with a heavy metal cation immobilized thereon to capture $A\beta$ peptide on the surface of the solid support, thereby forming a first complex which comprises solid support/heavy metal cation/A\beta peptide;
- blocking all exposed metal binding sites remaining after $A\beta$ capture with a blocker:
- (c) contacting the first complex, which has been passed through step (b), with an antibody specific for $A\beta$ peptide to form a second complex which comprises solid support/heavy metal cation/AB peptide/ antibody specific for $A\beta$ peptide;
- (d) labelling the second complex to form a detectable third complex which comprises solid support/heavy metal cation/A β peptide/ antibody specific for $A\beta$ peptide/label; and
- (e) detecting the third complex, and quantifying $A\beta$ peptide which may be present in the candidate solution.
- A diagnostic assay for detecting and/or quantifying $A\beta$ peptide which may be present in a candidate solution, comprising:
- (a) contacting the candidate solution with a solid support with a heavy metal cation immobilized thereon to capture $A\beta$ peptide on the surface of the solid support, thereby forming a first complex which comprises solid support/heavy metal cation/Aβ peptide;
- (b) blocking all exposed metal binding sites remaining after $A\beta$ capture with a blocker;
- contacting the first complex, which has been passed (c) through step (b), with an antibody specific for $A\beta$ peptide, called $A\beta$ antibody, to form a second complex which comprises solid support/heavy metal cation/A β peptide/A β antibody;

30

- (d) contacting said second complex with one or more antiantibodies specific to the $A\beta$ antibody to form a third complex which comprises solid support/heavy metal cation/ $A\beta$ peptide/ $A\beta$ antibody/one or more anti-antibodies;
- (e) labelling said third complex to form a detectable fourth complex which comprises solid support/heavy metal cation/A β peptide/A β antibody/one or more anti-antibodies/label; and
- (f) detecting the fourth complex, thereby quantifying $A\beta$ peptide which may be present in the candidate solution.
- 3. A diagnostic assay as claimed in claim 1, wherein said heavy metal cation is selected from the group consisting of zinc (II) and copper (II) complexed to nitriloacetic acid.
- 4. A diagnostic assay as claimed in claim 2, wherein said heavy metal cation is selected from the group consisting of zinc (II) and copper (II) complexed to nitriloacetic acid.

5. A diagnostic assay as claimed in claim 3, wherein said antibody at step (c) is specific to $A\beta_{1-42}$ and does not cross react with $A\beta_{1-40}$.

The

6. A diagnostic assay as claimed in claim 3, wherein said antibody at step (c) is specific to $A\beta_{1.40}$ and does not cross react with $A\beta_{1.42}$.

The

7. A diagnostic assay as claimed in claim 4, wherein said antibody at step (c) is specific to $A\beta_{1.42}$ and does not cross react with $A\beta_{1.40}$.

The

8. A diagnostic assay as claimed in claim 4, wherein said antibody at step (c) is specific to $A\beta_{140}$ and does not cross react with $A\beta_{142}$.

The

9. A diagnostie assay as claimed in claim 5, wherein said antibody is labelled with a radioisotope.

10

5

15

25

10

10. A diagnostic assay as claimed in claim 6, wherein said antibody is labelled with a radioisotope.

The

- 11. A diagnostic assay as claimed in claim 7, wherein said antibody is labelled with a radioisotope.
- 12. A diagnostic assay as claimed in claim 8, wherein said antibody is labelled with a radioisotope.
- 13. A diagnostic assay as claimed in claim 5, wherein said enzyme is horseradish peroxidase.
- 14. A diagnostic assay as claimed in claim 6, wherein said enzyme is horseradish peroxidase.

The

- 15. A diagnostic-assay as claimed in claim 7, wherein said enzyme is horseradish peroxidase.
- 16. A diagnostic assay as claimed in claim 8, wherein said enzyme is horseradish peroxidase.
- 17. A kit for carrying out the assay of claim 1 or 2, which comprises a carrier means compartmentalized in close confinement therein to receive one or more container means which comprises a first container means containing a solid support having a heavy metal cation immobilized thereon and a second container means containing an antibody specific for $A\beta$ peptide.

18. A kit as claimed in claim 17, wherein said heavy metal cation is selected from the group consisting of zinc (II) and copper (II) complexed to nitriloacetic acid.

20

18

- 19. A kit as claimed in claim 17, wherein said antibody is labelled with a radioisotope.
- 20. kit as claimed in claim 17, wherein said enzyme is whorseradish peroxidase.
- 21. A kit as claimed in claim 17, wherein said carrier means further comprises a third container means containing an anti-antibody which is specific for the $A\beta$ antibody.
- 22. A kit as claimed in claim 21, wherein said anti-antibody is labelled with a radioisotope.
- 23. A kit for carrying out the assay of claim 1 or 2, which comprises a carrier means compartmentalized in close confinement therein to receive one or more container means which comprises a first container means containing a solid support having a heavy metal cation immobilized thereon and a second container means containing a labelled antibody specific for $A\beta$ peptide.
- 24. A kit as claimed in claim 23, wherein said heavy metal cation is selected from the group consisting of zinc (II) and copper (II) complexed to nitriloacetic acid.
- 25. A kit as claimed in claim 23, wherein the labelled antibody is labelled by a radioisotope.
- 26. Kit as claimed in claim 23, wherein said enzyme is horseradish peroxidase.
- 27. A kit for carrying out the assay of claim 1 or 2, which comprises a carrier means compartmentalized in close confinement therein to

20

receive one or more container means which comprises a first container means containing a solid support having a heavy metal cation immobilized thereon and a second container means containing an antibody specific for $A\beta$ peptide bound to a labelled anti-antibody.

5

10

A kit as claimed in claim 27, wherein said heavy metal cation is selected from the group consisting of zinc (II) and copper (II) complexed to nitriloacetic acid.

28

29

kit as claimed in claim 27, wherein the labelled antibody is labelled by a radioisotope.

30. kit as claimed in claim 27, wherein said enzyme is horseradish peroxidase.

 \mathbb{A} method for purification of $\mathbb{A}\beta$ peptide from biological fluids

which comprises:

(a)

methylating cysteine groups of peptides in the biological

fluid:

acidifying the biological fluid obtained from step (a); (b)

applying the biological fluid obtained from step (b) to (c) a copper-charged chelating-Sepharose column;

(d) washing the column with equilibration buffer to obtain an eluate solution; and

(e) collecting the eluate solution, thereby obtaining purified $A\beta$ peptide.

A method for purification of $A\beta$ peptide from biological fluids which comprises:

(a) methylating cysteine groups of proteins in the biological fluid;

> acidifying the biological fluid obtained from step (a); (b)

20

15

- adding to the biological fluid obtained from step (b), a (c) free copper-charged chelating slurry to form a mixture;
- (d) centrifuging the mixture obtained from step (c) to obtain a pellet;
- washing the pellet obtained from step (d) with (e) equilibration buffer, thereby obtaining purified $A\beta$ peptide.
- A kit for carrying out the assay of claim 31 which comprises 33. a carrier means compartmentalized in close confinement therein to receive one or more container means which comprises a first container means containing a copper charged chelating-Sepharose column and a second container means containing an antibody specific for $A\beta$ pertide which may be used to confirm presence of purified $A\beta$ peptide.
- 34. A kit for carrying out the assay of claim 32 which comprises a carrier means compartmentalized in close confinement therein to receive one or more container means which comprises a first containing free copper-charged chelating-Sepharose and a second container means containing an antibody specific for $A\beta$ peptide which may be used to confirm presence of purified $A\beta$ peptide.

5

Hart of the Hart of the Hart Hart Hart Hart Hart ± 15 Hard then made